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ASCORBIC ACID STATUS IN PREGNANCY AND POSTPARTUM WOMEN IN ENUGU, SOUTH-EAST, NIGERIA: ARE VITAMIN SUPPLEMENTS SUFFICIENT?

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ABSTRACT

Pregnancy has been shown to be a period of stress, during which the nutritional needs of the foetus is dependent on that of the mother. It is also a condition exhibiting increased susceptibility to oxidative stress, leading to potential damage. Ascorbic acid is a strong antioxidant as well as a reducing agent and is increasingly utilized to normalize the pro-oxidant - antioxidant balance during pregnancy. Since ascorbic acid cannot be synthesized by the body, leading to total dependence on exogenous supply in diets, there is need to evaluate the level of the vitamin in pregnant women and determine their predisposition or otherwise to avitaminosis C. Ascorbic acid levels of 60 apparently healthy pregnant women, aged between 20-40 years old and attending the antenatal clinic at the UNTH, Enugu, 60 age-matched women in their postpartum and 60 age-matched non-pregnant women (controls) were assayed using the 2,4-dinitrophenyl hydrazine method. The serum ascorbic acid levels were significantly lower ($P<0.05$) in pregnant women, showing a steady decline with gestational age increase. There were statistically significant decreases ($P<0.05$) in the second and third trimesters whereas a significant increase ($P<0.05$) was observed in the postpartum period, when compared to the controls. The serum levels of ascorbic acid were decreased significantly as pregnancy progresses; in spite of the supplements given Significant increases in the postpartum period may be attributed to the absence of o xidative stress and recovery from stress. Additional supplementation and increased dietary intake should be encouraged in pregnant women.

Key words: Pregnancy, vitamin C, trimesters, supplementation,

INTRODUCTION

Pregnant women have been known to have high demand for nutrients during the different stages of their reproductive cycles (Ibeziako and Emitte, 1981). This increase in demand cuts across all nutrient classes. However, the most significant ones are those that can easily be lacking in our daily food, either through the preparation, processing and storage of food stuff (Yoshika, 1999). From early pregnancy, the human placenta influences maternal homeostasis, hence increased susceptibility to oxidative stress; a disturbance in the pro-oxidant-antioxidant balance, in favor of the former, leading to potential damage (Bourn, 2002). Also, during pregnancy, there are dynamic changes in multiple body systems resulting in increased basal oxygen consumption and also in changes in energy substrate use by different organs including the foeto-placental unit (MorieraSodre, 2003; Awoyelu et al, 2004).

Ascorbic acid is a water-soluble vitamin as well as a strong anti oxidant and a reducing agent necessary for the prevention of peroxidation reactions activated by reactive species generated in the course of intermediary metabolism (Krishma and Venkatarama, 2007). There is increasing evidence that there is increased vitamin requirement during pregnancy and since human beings are unable to synthesize ascorbic acid, they depend solely on endogenous supply of the vitamin in diet (Hemma, 2003).

Various functions have been ascribed to vitamin C, including acting as a reducing agent in biosynthesis, potentiating the action of enzymes and other vitamins, facilitating

absorption of ferrous sulphate and protection against bacterial infections, especially acute respiratory infections (Khalil and Waly, 1989; Ibadin et al, 1999). Due to the diverse functions of the vitamin, there is need to evaluate the ascorbic acid levels in the different trimesters of pregnant women on ascorbic acid supplementation and also in the postpartum women to ascertain if the supplements given during pregnancy is adequate or not.

MATERIAL AND METHODS

Subjects: The study is a cross sectional study of apparently healthy pregnant women on vitamin C supplement and those in the post partum period attending the antenatal care clinic in the maternity wing of the University of Nigeria Teaching Hospital, UNTH, Enugu. The subjects were aged between 20-40 years old, and were divided into four groups based on their gestational trimesters and post partum period. 60 subjects were recruited from each trimester of their pregnancy whereas 60 were in their post partum period. Sixty (60) apparently healthy, age-matched, non-pregnant women, comprising of the students and staff of UNTH, Enugu served as the control subjects. Informed consent was given by all the subjects and ethical clearance was issued by relevant authorities before the commencement of the study.

Exclusion Criteria: Subjects with pregnancy complications like gestational diabetes, hypertension, pre-eclampsia, asthma, anemia and jaundice were excluded from the study.

Sample Collection and Processing

Venous blood samples (5ml) were collected in clean plain tubes and were allowed to clot and retract at room temperature. The samples were then centrifuged at 5000 rpm and the clear serum samples were promptly separated into plain tubes.

The samples were analyzed immediately for ascorbic acid.

Analytical Method

Ascorbic acid was assayed using the 2,4-dinitrophenyl hydrazine (2,4-DNP) method of Caraway (Tietz, 1996). Oxidized ascorbic acid was reacted with 2, 4-DNP, 10% TCA and chloroform in a strong acidic medium (85% sulphuric acid) and the red colouration generated was estimated at 490nm using Spectronic 20 spectrophotometer.

Statistical Analysis:

The statistical analysis was done using two-tailed students t-test and the results are presented as mean ± standard deviation (± SD) (Ojo, 1995).

RESULT

Table 1 shows the mean ± SD of ascorbic acid levels in different trimesters postpartum and controls. From the table, the ascorbic acid levels decreased with increasing gestational age and increased at the post partum period.

Subjects	Number	Ascorbic Acid (mg/100ml)
First Trimestrer	n=60	0.98 ± 0.16
Second Trimester	n=60	0.93 ± 0.15
Third Trimester	n=60	0.88 ± 0.11
Postpartum	n=60	1.15 ± 0.17
Control	n=60	1.04 ± 0.21

When compared with the control, the reduction at the second and third trimesters as well as the increased level observed in the post partum were statistically significant ($P<0.05$) (table 2).

TABLE 2

Test of difference in means of ascorbic acid between control, postpartum period and the different trimesters.

Subjects	Ascorbic Acid (mg/100ml)
First Trimester and Control	P> 0.05
Second Trimester and Control	P< 0.05 **
Third Trimester and Control	P< 0.05 **
Postpartum and Control	P< 0.05 **

** = Statistically significant

Table 3 however, shows the test of difference between the postpartum period and the different trimesters. The decrease in the mean ascorbic acid levels as pregnancy progressed was statistically significant ($P<0.05$) compared to the level observed in the post partum period.

TABLE 3.
Test of difference in means of ascorbic acid level between the postpartum period and the different trimesters of pregnancy.

Subjects (groups)	Ascorbic Acid (mg/100ml)
First Trimester and Postpartum	P<0.05 **
Second Trimester and Postpartum	P<0.05 **
Third Trimester and Postpartum	P<0.05 **

** = Statistically significant

DISCUSSION

Vitamin C has many vital functions in the body. Therefore, it is important to have a way of measuring it (Jackson et al, 2005). The present study evaluated the level of vitamin C in the different trimesters of pregnancy. The study revealed a progressive fall in the ascorbic concentration with increasing gestational age. There was statistically significant decrease (P<0.05) in the second and third trimesters of pregnancy and also a significant increase (P<0.05) in the postpartum period when compared with the control subjects.

The significant decrease in the second and third trimesters could be as a result of stress secondary to pregnancy in addition to increased fetal demand (Awoyelu et al, 2004), and if not properly addressed, the pregnant women as well as the developing fetus may be deprived of the very essential functions of vitamin C in the body.

Ascorbic acid is involved in the metabolism of several amino acids, leading to the formation of hydroxyproline, hydroxylysine, norepinephrine, serotonin, homogenistic acid, and carnitine (Barnes, 1975). Hydroxyproline and hydroxylysine are components of collagens, the fibrous connective tissue in animals. Collagens are principal components of tendons, ligaments, skin, bone, teeth, cartilage, heart valves, intervertebral disks, cornea, eye lens, and the ground substances between cells (Hacisevk, 2009). When collagen is synthesized, proline and lysine are hydroxylated post-translationally on the growing polypeptide chain. Hydroxyproline and hydroxylysine are required for the formation of a stable extracellular matrix and cross-links in the fibers (Hacisevk, 2009).

In a variety of other functions, the role of ascorbic acid in cellular metabolism can be accounted for by its reducing properties to protect cellular components from oxidative damage. It acts as a scavenger for oxidizing free radicals and harmful oxygen-derived species, such as the hydroxyl radical, hydrogen peroxide, and singlet oxygen (Arrigoni and Tulio, 2002). Since ascorbic acid is implicated in synthesis of collagen which is in turn a principal component of tendons, ligaments, skin, bone etc (Hacisevk, 2009), the development of the fetus will be adversely be affected if the requirement for ascorbic acid is not met by the mother. Hence, supplementation remains the proper approach to ensure that adequate requirements are met.

The half-life of vitamin C in tissues varies depending on the literature data, from 16 to 20 days. Assuming a tissue level of 5000 mg, a lack of vitamin C in the diet for 16 days would reduce the tissue store to about 2500 mg; in 32 days it would be about 1250 mg; in 44 days it would be 625 mg and in 64 days it would be about 313 mg and clinical signs of scurvy should start to develop (Sauberlich, 1985). Vitamin C is usually available in fresh fruits and vegetables, especially citrus fruits and expectant mothers are expected to consume a reasonable amount to maintain the required dietary recommendation, but most times people prefer to go for already packaged fruit juice, with lots of preservatives and little or no vitamin C.

The significant increase in postpartum period may be attributed to absence of oxidative stress, increased consumption and subsequent recovery from stress. Ascorbic acid is water-soluble and is well absorbed from the gastrointestinal tract. Mean plasma ascorbic acid levels are 50-60 μM for healthy, well-nourished, non-smoking individuals (Duarte and Lunec, 2005). Plasma levels can be increased by long-term vegetarian diet (Astley, 2004) and by oral supplementation up to approximately 100 μM (Choi et al, 2004). Higher plasma levels are not observed even with supplemental doses higher than 500 mg/day due to efficient vitamin C excretion in the urine (Hacisevk, 2009). Some studies have shown that the increase in plasma vitamin C was accompanied by an increase in the intracellular

However, this increase is often not dose-dependent (Duarte and Lunec, 2005), presumably due to cellular saturation. Thus it is known that the intracellular vitamin C concentrations of neutrophils, monocytes and lymphocytes saturate at lower supplementation doses than human plasma (Hacisevk, 2009).

Even when dietary intake is adequate, pregnancy usually induce increased requirement of ascorbic acid. The amount of vitamin required by an individual has been shown to vary considerably, and may be influenced by such factors as body size, growth rate, physical activity and pregnancy. Oral vitamin C supplementation has been reported to lead to correction of haemostatic dysfunction resulting from chronic smoking (Soronnadi et al, 2013). The recommended dietary daily allowances for vitamin C are 90 mg for men and 75 mg for women. At intakes of the vitamin about 60 mg/d in both genders, ascorbate begins to appear in the urine. However, intakes of 250 mg/d and higher are required to saturate ascorbate concentrations in plasma and contents of white blood cells (Levine et al, 2001).

Irrespective of the fact that 300mg of ascorbic acid is given as part of routine antenatal drug supplementation; significant reduction was still recorded in the level of the vitamin in pregnant women. This goes to show that the vitamin C supplementation for pregnant women in our environment is insufficient. There should be further study to determine the actual dose that will be appropriate

CONCLUSION

The enormous benefits of adequate serum levels of ascorbic acid during pregnancy cannot be over emphasized. Having ascertained, from the study, that serum levels of ascorbic acid were significantly lowered as pregnancy progresses in spite of the supplements given, efforts should therefore be made to encourage the consumption of vitamin C-rich diets such as citrus fruits and vegetables pregnant women. Other antioxidants like vitamin E may also be encouraged. This will go a long way in reducing maternal and pre-natal mortality and also other likely complications that may result from vitamin C deficiency.

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